Vol. XL. *March, 1921.* *No. 3*

BIOLOGICAL BULLETIN

DIFFERENCES IN VIABILITY IN DIFFERENT TYPES OF REGENERATES FROM DISSOCIATED SPONGES, WITH A NOTE ON THE ENTRY OF SOMATIC CELLS BY SPERMATOZOA.

JULIAN S. HUXLEY,

NEW COLLEGE, OXFORD.

Having occasion recently to go over the notes on which a previous paper of mine[1] was based, I came across one result which, although not published at the time, now seems worthy of record.

In the paper referred to, it was shown that by filtering chopped Sycons through coarse instead of fine gauze, and pipetting off the first-deposited portion of the cell-sediment, masses of cells could be produced consisting entirely or almost entirely of choanocytes.[2] The unpublished data concern the occurrence, in cultures of such collar-cell masses, of apparently normal regenerating masses similar to those obtained by straining through fine gauze.[3] This would appear to indicate that the chemotactic or other attraction exerted by amœbocytes and dermal cells upon each other is stronger than the similar attraction exerted upon these same cells by choanocytes.[4] This would lead to the observed differential separation of the bulk of the dermal and amœboid cells in preparations where they were present only in very small relative number.

These *normal regenerates,* as they may be called, in opposition to *collar-cell masses,* were found in four of my twelve cultures

1 Huxley, *Phil. Trans. Roy. Soc.* (*B*), Vol. 202, 1911.
2 *Ibid.*, p. 177.
3 *Ibid.*, pp. 167–170.
4 *Ibid.*, p. 167.

of collar-cell masses. In every case, they lived longer than the collar-cell masses.

The time that elapsed before all the collar-cell masses (whether spheres or blown-out masses) in any one of these four cultures had shown the first sign of impending death by contracting, was from 6 to 12 days; the time before all the masses of a culture were dead, from 8 to 15 days. The time which elapsed before the normal regenerates in the same culture died, however, was from 14 to 33 days; in most cases, all normal regenerates lived longer than any collar-cell mass in the same culture. Whether this greater viability of the masses containing all kinds of cells in approximately normal proportions was due to a protective function exerted by the dermal cells after their migration to the exterior, or to the fact of some dermal or amœboid cells serving as food for the rest, or to other possible causes, remains to be seen. In any case, the facts are interesting.

A further observation may be referred to. It appears that the spermatozoa of calcareous sponges have very rarely been observed. Dr. Gatenby, of University College, London, who has been working on the fertilization of sponges, was discussing the subject with me, when I recalled that bodies resembling spermatozoa had been visible in some of my preparations of normal regenerating masses from dissociations.

Some of my slides I lent to Dr. Gatenby, who re-stained them, and was thus able to discover certain interesting facts. The facts are briefly as follows: Round the margin of all cell-masses from some of my experiments are to be seen minute deeply-staining bodies resembling spermatozoa with an ordinary elongated head; and groups of such bodies are usually to be seen in the interior of the preparations. They are, however, totally absent from other slides representing other experiments. There can be no doubt that these are spermatozoa. The interesting point about them is that they swarm round the masses of cells whether these contain oöcytes or not. *I.e.,* such sponge spermatozoa are attracted by somatic cells as well as by their proper partners.

On re-staining, Dr. Gatenby found in the interior of many of the somatic cells bodies which could be interpreted as heads of spermatozoa which had been half converted into vesicular nuclei.

Here, however, it is impossible to give full proof of this until further material can be examined. But it is at least suggestive that Dr. Gatenby himself[1] has found in the normal fertilization of a closely-related sponge that the spermatozoa do not penetrate the oöcytes directly, but enter collar-cells. Within these they undergo a partial transformation to vesicular nuclei (at this stage closely resembling the bodies found in the cells of the regenerating masses), and are then transferred, by the migration of the collar-cells, to the oöcytes, into whose substance they pass. Within the maturing ova they undergo the remainder of the transformation to male pronuclei, and then effect fertilization in the usual way. If the bodies within the cells of the regenerating masses do prove to be what they appear to be, namely, half-transformed sperm-heads, two interesting points emerge. The first is that somatic cells can exert an attraction on spermatozoa comparable to that exerted by oöcytes. In normal fertilization, only collar-cells within a certain radius are entered by spermatozoa; thus it might be supposed that the attraction was exerted by the oöcyte *through* the collar-cells, and that these had no attraction of their own. That this is not so, would be proved if our interpretation of the bodies in the regenerating masses is correct. But we would have to suppose that this attraction of the collar-cells was much less than that of the oöcytes, whose presence thus would prevent collar-cells beyond a certain distance from oöcytes from being entered.

In the second place, the definite but slight attraction of the collar-cells for spermatozoa would be correlated with the definite but partial transformation of the sperm-head to a nucleus within them. This correlation between degree of attraction and degree of nuclear transformation is what would be expected on such a theory of fertilization as Lillie's.

It is to be hoped that any worker having the opportunity to examine ripe sponge spermatozoa will undertake an investigation of the problem raised in this note.

In conclusion I have to thank Dr. Gatenby for his interest and for permission to publish the results discovered by him.

1 "The Cytoplasmic Inclusions of the Germ-Cells. Part VIII. Fertilization and Gametogenesis in *Grantia compressa*," *Journ. Linnaean Soc.*, 1920.

May 28, 1920.